Evaluation of Anti-Ulcer activity of Pseudarthria viscida linn *in Experimental animals*

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ABSTRACT

Objective: **To evaluate the antiulcer activity of *Pseudarthria Viscida* Linn. Root extract on experimental rats.**

Methods:

The antiulcer activity of PVRE was studied using ethanol (Absolute Ethanol) & indomethacin induced acute gastric ulcer models. The animals were divided into six groups; each group contains six animals. PVRE was administered in two doses, (200 mg/kg and 400 mg/kg, p.o) before 1 h to administration of ethanol & indomethacin. The parameters investigated include acid volume, pH, total acidity, ulcer index, MPO, LDH, lipid peroxidase, catalase & histopathological studies.

Results:

In antiulcer study, the extract significantly inhibited the development of ulcers induced by ethanol & indomethacin. The PVRE significantly reduced the acid volume, total acidity, total acidity, ulcer index, lipid peroxidation, LDH, MPO & increases in pH, & catalase level. Histopathogical studies also revealed that PVRE is gastro-protective. Pantoprazole (30 mg/kg) is used as standard drug.

Conclusion:

All the observation implies that PVRE possess significant protective activity against ethanol and indomethacin induced gastric ulcer in experimental rats. 400 mg/kg dose has shown more protection compared to 200mg/kg.

Keywords: **Antiulcer; *Pseudarthria Viscida* Linn.; Ethanol; Indomethacin; Pantoprazole**

INTRODUCTION

Peptic ulcer disease (PUD)

A peptic ulcer is a sore in the lining of the stomach or the first part of the small intestine. Ulcers in the stomach are called gastric ulcers, and those in the upper small intestine are called duodenal ulcers. Gastric and duodenal ulcers are breaks in the gastric and duodenal mucosa. Both gastric and duodenal ulcers relate to the corrosive action of pepsin and hydrochloric acid on the mucosa of the upper gastrointestinal tract. Ulcers generally range between 3 mm and several centimeters in diameter[1].

Peptic ulcers are usually aggravated by an imbalance between destructive and defensive factors in the stomach. The endogenous destructive factors in the stomach are HCl, pepsin, biliary reflux, lipid peroxidation, and the formation of reactive oxygen species (ROS) and the exogenous factors are excessive use of ethanol, indiscriminate use of non-steroidal anti-inflammatory drugs (NSAID), stress, smoking, and infection by Helicobacter pylori bacteria. The defensive factors are mucus-bicarbonate barrier, mucin secretion, surface phospholipids, prostaglandins (PGs), nitric oxide (NO), mucosal blood flow, cell renewal, growth factors, and antioxidant enzymes. Oxidative stress, present in the process of gastric ulceration, increases the formation of ROS that can disrupt epithelial cell integrity. An excess production of ROS metabolites may overwhelm the endogenous antioxidants. In addition, ROS accumulates neutrophils in the tissues of the mucosa during gastric ulceration. Studies have shown that pro-inflammatory cytokines induce the activation of neutrophils and are strong contributors to the of ulcer damage [2]. Gastric ulcers develop in the lining of the stomach. Pain from a gastric ulcer may be aggravated or alleviated by eating. Duodenal ulcers form in the first few centimeters of the lining of the duodenum (the upper part of the small intestine). Pain associated with duodenal ulcers is typically relieved by eating but often returns two to three hours later and frequently awakens patients at night[3].

ETIOLOGY OF PEPETIC ULCER

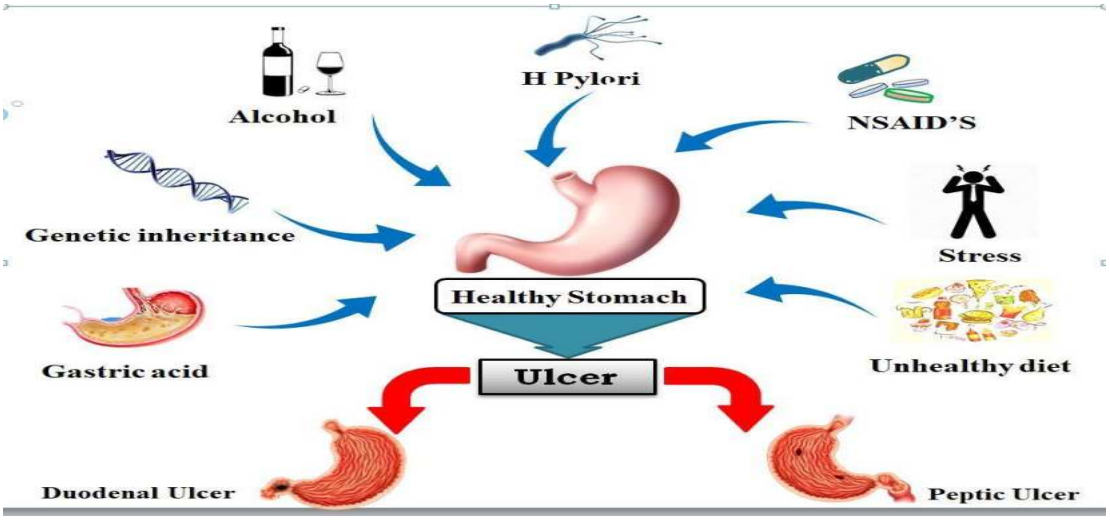


FIGURE: Etiology of Peptic Ulcer

Screening methods for Anti-ulcer activity

1. **Ethanol induced gastric lesions**[4]

Intragastric application of absolute ethanol is a reproducible method to produce gastric lesions in experimental animals. The protective effect against various irritants has been called

cytoprotective activity. Male Wistar rats weighing 200-250 g are deprived of food 24 h prior to the experiment but are allowed free access to water. The rats are administered either are appropriate vehicle or the cytoprotective drugs, intragastrically 30 min prior to administration of 1 ml absolute ethanol. 1 h after administration of ethanol, the animals were sacrificed under anesthesia using ether and their stomachs exercised, cut along the greater curvature and gently rinsed under tap water.

2. Indomethacin induced ulcers in rats[5]

NSAIDs agents, like indomethacin and acetyl-salicylic acid, induce gastric lesions in men and in experimental animals. Wistar rats weighing 150-200g are used. The test drugs are administered orally in 0.1% Tween 80 solution 10 min. prior to oral indomethacin in a dose of 60 mg/ kg 4 h.later, the animals were sacrificed under anesthesia using ether and their stomachs removed and opened along the greater curvature, washed in warm water, and examined under a 3-fold magnifier.

1. Pylorus ligation in rat (SHAY RAT)[6]

SHAY rat is a simple and reliable method for production of gastric ulceration in the rat based on ligature of the pylorus. The ulceration is caused by accumulation of acidic gastric juice in the stomach. Ulcer index and acidity of the gastric content of treated animals are compared with controls. Female Wistar rats weighing 150-170 g are starved for 48 h. having access to drinking water ad libitum. During this time, they are housed single in cages with raised bottoms of wide wire mesh in order to avoid cannibalism and coprophagy. Ten animals are used per dose and as controls. Under ether anesthesia a midline abdominal incision is made. The pylorus is legated; care being exercised that neither damage to the blood supply nor the pylorus occurs. Grasping the stomach with instruments is to be meticulously avoided; the abdominal wall is closed by sutures. The test compounds are given either orally by gavage or injected subcutaneously. The animals are placed for 19 h in plastic cylinders with an inner diameter of 45 mm being closed on both ends by wire mesh. Afterwards, the animals are sacrificed under ether anesthesia. The abdomen is opened and a ligature is placed around the esophagus close to the diaphragm. The stomach is removed, and the contents are drained in a centrifuge tube. Along the greater curvature the stomach is opened and pinned on a cork plate. The mucosa is examined with a stereomicroscope.

2. Stress ulcer through immobilization stress[7]

Stress plays a major role in the pathogenesis of gastric ulcers in man. The experimental model resembles the psychogenic factors in the pathogenesis of gastric ulcers in patients. Groups of 10 female Wistar rats per dose of test drug and for controls weighing 150- 170 g are used. Food and water are withdrawn 24 h before the experiment. After oral or subcutaneous administration of the test compound or the placebo solution the animal's extremities are fixed together and the animals are wrapped in wire gauze. They are horizontally suspended in the dark at 20°C for 24 h and finally sacrificed under ether anesthesia. The stomach is removed.

3. Stress ulcers by cold water immersion[8]

Cooling of rats in water during the restraint period accelerates the occurrence of gastric ulcers and shortens the time of necessary immobilization. Wistar rats weighing 150-200 g are used. After oral administration of the test compound, the rats are placed vertically individual restraint cages in water at 22°C for 1 h. Then, they are removed, dried and injected intravenously via the tail vein with 30 mg/ kg evans blue. Ten min. later, they are sacrificed under ether anesthesia and their stomach was removed. Formol - saline (2%v/v) is then injected into the totally ligated stomachs for storage overnight.

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7. Sub-acute gastric ulcer in rats[7]

This is a method for producing standard subacute gastric ulcers in rats and for the quantitative evaluation of the healing process. Female Wistar rats weighing 120-150 g are fasted for 24 h having access to water ad libitum in cages with wire sieves at the bottom. The rats are anesthetized with ether and a polyethylene catheter including a fine steel wire with a needle tip (1.2 mm diameter) at the lower end is orally inserted into the stomach. After the cannula reaches the gastric wall, the upper end of the steel wire is pressed in a definitive manner, so as to puncture the gastric wall. The test substances are administered orally, 30 min. prior or 24 h after puncture. Free access to food and water is provided from 2 h up to the end of the experiment. Each group consists of 8 - 15 rats. The animals are sacrificed by overdose of ether at definitive time intervals after puncture. The stomach is dissected and opened along the lesser curvature.

8. Aspirin induced ulcer genesis in pylorus ligated rats[9]

In aspirin-induced ulcerogenesis in pylorus ligated rats, aspirin is administered at a dose of 200mg/kg orally in a suspension prepared in 1% CMC with water 1h prior to pyloric ligation (time interval between reference drugs and aspirin should be 1h) and the process described above was followed.

9. Acetic acid-induced gastric ulcers[10]

Rats were starved for 24 h prior to the experiment and were divided into four groups, one for control and the other three for drug treatment. Under light ether anaesthesia, laparotomy was performed through a midline gastric incision. After exposing the stomach, 0.05 ml of 10% v/v acetic acid was injected into the subserosal layer in the glandular part of the anterior wall. The stomach was bathed with saline to prevent adhesion to the external surface of the ulcerated region. The vehicle, extract and standard drug was administered orally for 20 days beginning One day after surgery. On the 21st day the animals were sacrificed at proper intervals to assess the healing processes of the ulcer. The stomach was removed and the gastric lesions were evaluated by examining the inner gastric surface with a dissecting binocular microscope. Subsequently the ulcer area (mm) and curative rate % were determined.

10. Cysteamine-induced duodenal ulcers[7]

The method described by Szabo (1978) was followed. Duodenal ulcers were induced by administration of two doses of cysteamine hydrochloride, 400 mg /kg, *p.o.* in 10% aqueous solution at an interval of 4 h. use at dose levels of 100 and 200 mg/kg rantidine (50mg/kg, *p.o.*) wre administered 30 min before each dose of cysteamine hydrochloride. All the animals were sacrificed 24 h after the first dose of cysteamine and dudodena were excised carefully and opened along the antimesentric side.

10. Dimaprit induced gastric ulcers [9]

On the basis of universally recognised hypothesis about the involvement of H_2 receptors in the pathogenesis of peptic ulcer an appropriate rat model has been designed with dimaprit as scientific H_2 receptor agonist. Dimaprit was administered *i.p.* or *i.v.* to 24 h fasted rats and the animals were sacrificed 4 h after the injection. The drugs for studying their gastroprotective effects were given 30 min before dimaprit.

PLANT PROFILE

Plant Name: ***Pseudarthria Viscida Linn.***

Common name: kalasi, prsniparni, salaparni, antubelegida, antuparni, muvila.

TAXONOMICAL CLASSIFICATION:

Plant name: ***Pseudarthria viscida*** **Linn.**

Kingdom: Plantae

Order : Fabales

Family : Fabacea

Genus : *Viscida*

Species : *P. viscida.*



FIGURE1: *PSIUDARTHRIA VISCIDA* PLANT

MORPHOLOGY OF *PSEUDARTHRIA VISCIDA*

Pseudarthria viscida **(Fabaceae) is a shrub a semi erect, pubescent, perennial shrub grows up to 50 cm to 1 meter in height. Branches are covered with whitish hairs. Leaves are hairy, compound; trifoliate, terminal leaflet rhomboid ovate, lateral ones are obliquely ovate or oblong. Flowers are purplish or pink, small numerous, found on terminal and axillary spikes. Fruits pods, flattened and covered with hairs, seeds 4-6[2]**

DISTRIBUTION

Globally the species is distributed in the Indo-Malaysian region and Sri Lanka. In India it is recorded in the states of Gujarat, Orissa, Karnataka, Tamil Nadu and Kerala.

ACTIVE CONSTITUENTS

Alkaloids, glycosides, carbohydrates, proteins, steroids, flavonoids, terpenoids, tannins & phenolic compounds (Gallic acid, Caffeic acid, Rutin, Quercetin and Ferulic acid) and fixed oils [11]

Medical use

The roots are astringent, sweet, bitter, emollient, digestive, constipating, anthelminthic, cardiotonic, febrifuge and tonic. They are useful in vitiated conditions of cough, bronchitis, asthma, tuberculosis, helminthiasis, diarrhoea, inflammation, cardiopathy, fever, haemorrhoids, gout, diabetes, hyperthermia and general debility

Reported activities[12]

- **Antifungal activity**
- **Anti-inflammatory activity**
- **Analgesic activity**
- **Anti-diabetic activity**
- **Anti – microbial activity**
- **Anti –oxidant activity**

MATERIALS AND METHODS

Experimental Animals.

Experimental animals were procured from vivarium of Sree Siddaganga college of pharmacy, Tumakuru, Karnataka and maintained with 12hr light: 12h dark normal cycle study protocol was approved by institutional animal ethics committee (IAEC)of Siddaganga college of pharmacy, Tumakuru, Karnataka approval number SSCP/IAEC. Clear/199/2019-20. All the animals were acclimatized for 7 days prior to initiation of the experiment. Selected animals were marked individually, randomized into experimental and control group according to body weight distribution. So that mean body weight of each group would not be statistically different from those of the other groups. Animals were housed in sanitized polypropylene cages containing sterile paddy husk as bedding.

METHODS

Collection of plant material.

The roots of the *Pseudarthria viscida Linn* were collected from the local region of Tumakuru, Karnataka. The collected samples identified and authenticated by prof P. Chidananda Department of botany Tumakuru. And specimen voucher (herbarium) is preserved in the department.

Preparation of Hydro alcoholic extract of *Pseudarthria viscida Linn*

The roots of Pseudarthria viscida were shade dried until the roots become brittle. Later the roots were grounded into course powder and subject into Soxhlet extraction process using ethanol and water (70:30) as solvent the hot extraction was carried out using Soxhlet apparatus. After the process solvent obtained was dried using water bath and a semisolid residue of ethanolic extract of *Pseudarthria viscida Linn* was obtained.

PREPARATION OF EXTRACT AND TEST DRUG

1. **Hydro alcoholic extract of Pseudarthria viscida**

The Hydro alcoholic *Pseudarthria viscida Linn* root extract (PVRE) was dissolved in 0.3% w/v sodium CMC (carboxyl methyl cellulose) and administered orally to the experimentally animals.

2. **Preparation of ethanol**

Absolute ethanol at a dose of 5ml / kg used as inducer and administered orally to the experimental animals.

3. **Preparation of indomethacin**

Indomethacin (80mg/kg) solution was prepared in 0.3% CMC solution used as inducer drug in the study and administered orally to the experimental animals.

4. **Preparation of pantoprazole**

Pantoprazole (30mg/kg) solution was prepared in 0.3% CMC solution used as Standard drug in the study and administered orally to the experimental animals.

Statistical analysis

The data were expressed as mean ± SEM. The statistical analysis of data was done by using One-way of variance (ANOVA), followed by Turkey's multiple comparison test was used using graph pad prism 5.0 software (Graph pad, San Diego, CA)

1. **Pharmacological evaluation**

Pharmacological screening can be done to evaluate and achieve the desired therapeutic effect of particular drug able candidate. Also these screening methods allow us to understand the complete pharmacological effect through various bio- analytical processes. These studies afford the safety profile during complete investigations. To determine the effects of PVRE on experimentally induced gastric ulcer in rats were assessed with the help of following models:

a) **Ethanol induced gastric ulcer model**
b) **Indomethacin induced gastric ulcer model**

a). Ethanol induced Gastric ulcer model[5]

Female Wistar rats weighing (200-230g) were used for the study. Animals were randomized according to body weight and grouped into 6 groups as follows (n=6).

	Treatment
	Normal Control (vehicle)
	PVRE (400mg/kg)
	Ethanol (5ml/kg)
	PVRE (200mg/kg) + Ethanol (5ml/kg)
	PVRE (400mg/kg) + Ethanol (5ml/kg)
	Pantoprazole(30mg/kg) + Ethanol (5ml/kg)

Animals were pre-treated for 7 days. Animals were fasted for 24 - 48 hrs, prior to induction. On the 7th day after the 1hr treatment with PVRE and pantoprazole animals were induced with ethanol (5 ml/Kg). After 1 hour of induction animals were scarificed. Before the sacrification the blood was collected through retro orbital puncture and serum was separated for the estimation of LDH. Then the animals were scarified under euthanasia procedure. The abdomen were cut opened with clamping of pylorus to collect the gastric guice then stomach were removed cut along the greater curvature and spread to score ulcer index. The collected gastric juices were used for the analysis of pH and total acidity. After scoring the stomach were homogenized in a suitable buffer and used to estimate total protein, catalase, reduced glutathione (GSH), lipid peroxidation (LPO), lactate dehydrogenase (LDH), Myeloperoxidase (MPO), and histopathological studies.

b). Indomethacin induced gastric ulcer models [13]

Wistar rats of either sex (180-200g) were used for the study. Animals were randomized according to body weight and grouped into 6 groups as follows (n=6).

roups	Treatment
	Normal Control (vehicle)
	PVRE alone (400mg/kg)
I	Indomethacin (80mg/kg)
7	PVRE(200mg/kg) + Indomethacin (80mg/kg)
	PVRE(400mg/kg) + Indomethacin (80mg/kg)
I	Pantoprazole (30mg/kg) + indomethacin (80mg/kg)

Animals were pre-treated for 7 days. Animals were fasted for 24 - 48 hrs, prior to induction. On the 7th day after the 1hr treatment with PVRE and pantoprazole animals were induced with indomethacin (80mg/kg). After 4 hour of induction animals were sacrificed. Before the scarification the blood was collected through retro orbital method.

puncture and serum was separated for the estimation of LDH. Then the animals were scarified under euthanasia procedure. The abdomen were cut opened with clamping of pylorus to collect the gastric guice then stomach were removed cut along the greater curvature and spread to score ulcer index. The collected gastric juices were used for the analysis of pH and total acidity. After scoring the stomach were homogenized in a suitable buffer and used to estimate total protein, catalase, reduced glutathione (GSH), lipid peroxidation (LPO), lactate dehydrogenase (LDH), Myeloperoxidase (MPO), and histopathological studies.

Ulcer index[14]

The stomach were cut along greater curvature and flushed with saline to remove gastric content and blood clots. The stomachs were spread on the white thermacoal. The macroscopic evaluation and severity of the ulcer was assessed based on the clinical features of the stomach using a score scale ranging from 0-3 as follows.

Particulars	Score
Normal Stomach	0
Red Coloration	0.5
Spot ulcer	1
Haemorrhagic Streak	1.5
Ulcers	2
Perforations	3

Mean Ulcer score for each animal will be expressed as ulcer index

$$\%\ of\ ulcer\ protection = \frac{\text{control mean ulcer index} - \text{test mean ulcer index}}{\text{control mean ulcer index}} \times 100$$

Evaluation of gastric juice parameters

The stomach was cut along the greater curvature and the gastric content was collected by flushing 1ml of saline. After collection of the gastric content it was drained into the graduated centrifuge tube and tubes were centrifuged at 3500 rpm for 15 minutes. Then it was used for gastric juice analysis.

pH analysis [15]

The supernatant of the centrifuged gastric juice was analysed for the pH using pH strip.

Evaluation of total acidity [14]

Reagent: 0.1N NaOH was prepared by dissolving 0.4g of NaOH into 100 ml of distilled water The burette was rinsed and filled with 0.1 N NaOH. One ml of supernatant from the centrifuged tube was pipetted into 100 ml conical flask, two or three drops of phenolphthalein indicator were added and were titrated with 0.1N NaOH until the colourless solution turns to red tinge. The red colour tinge indicates the end point of the titration which corresponds to the total acidity. Acidity was calculated using the following formula.

$$Acidity = \frac{volume\ of\ NaOH \times Normality\ of\ NaOH \times 100}{0.1} \quad meq/L$$

Biochemical estimation

Tissue Preparation

1. **After macroscopical analysis of ulcer the stomach tissue samples were blotted to remove wet content and weighed.**
2. **Then the samples were chopped and homogenized at 3500 rpm in an ice cold potassium phosphate buffer (0.1M pH 7.4) 10% w/v.**
   0.1M phosphate buffer pH7.4 (6.059g of K_2HPO_4, 2.07g of KH_2PO_4)
3. **The suspension was centrifuged at 6000 rpm for 30 minutes. The supernatant was used for the estimation of LPO, GSH, CAT and Total protein. Portion of inflamed tissue was separately assayed for MPO quantification.**

Estimation of catalase [16]

Principal: - catalase is the second most abundant enzymatic antioxidant (after superoxide dismutase). It is an intracellular enzyme which dissociates Hydrogen peroxide (H2O2) into molecular O2 and Water (H2O2). This enzyme attenuates the level of reactive oxygen free radical or Reactive oxygen species (ROS). This conversion can be measured at 240 nm wavelength.

Reagents: -

1. **60mM Phosphate buffer (pH 7.4) :- 1.213 g of Disodium hydrogen Orthophosphate ($Na_2HPO_4.7H_2O$) and 0.204 g of sodium Dihydrogen orthophosphate ($NaH_2PO4.H_2O$) are dissolved in 80 ml of distil water and the desired pH is adjusted using HCl or NaOH. then the solution is made upto 100 ml using distil water.**
2. **10mM Hydrogen peroxide: - Add 0.58 ml of H2O2 (6%w/v) in 100ml of 60mM phosphate buffer**

Procedure

1. **Briefly, 0.95ml of 10mM of H2O2 in 60mM phosphate buffer (Ph7.0) was mixed with 50µl of tissue homogenate.**
2. **The amount of rate of degradation of H2O2 was measured spectrophotometric –ally at 240nm per minute.**

Calculation: -

Catalase content was expressed in terms of U/mg of tissue using formula

$$K=\frac{2.303}{\Delta t} \times Log \frac{A1}{A2} S\text{-}1$$

Estimation of LPO [17]

The assay is based on the reaction of thiobarbituric acid (TBA) with Malondialdehyde (MDA), a breakdown product derived from many oxidized lipid molecules. The resulting chromophore of TBA – MDA complex formation, which was measured at its absorbance maximum 532 nm

Reagents

TCA-TBA-HCl reagent

Solution 1 (15% w/v TCA): add 3g of TCA to 2 ml of TCA-TBA-HCl reagents and heat at 90 c for one hour at water bath.
Solution 2 (0.375% w/v TBA): add 75 mg of thiobarbituric acid (TBA) to 20ml of distil water Solution 3 (0.25 N HCl): add 0.43 ml of concentrated HCl in 20ml of distil water then mix all the solutions.

Procedure

1. **To 0.1 ml of tissue homogenate add 2ml of TCA- TBA-HCl reagent and heat at 90 for one-hour water bath.**
2. **Then cool the solution at ice-cold water, then centrifuge the mixture at 5500 rpm for 5 min and read the absorbance of clear supernatant at 535 nm**

Calculation

ε=1.56×10^5$M^{-1}cm^{-1}$ and the results were expressed as µM/g of tissue.

Estimation of Lactate dehydrogenase-P

Collected blood samples were kept in a non EDTA coated tubes and allowed for clotting for a period of 45 minutes. Later, blood samples were centrifuged at 3000 rpm at 4 c to get a clear serum. Obtained serum was used for estimation of lactate dehydrogenase-P using ERBA Diagnostic kit Germany (DGKC Method, kinetics)

Clinical significance

Lactate dehydrogenase is a cytoplasmic cellular enzyme presents in lysosomes of most tissues and organs that catalyses the interconversion of pyruvate to lactate which is the final product of death cell glycolysis. Serum analysis of LDH used for detection of cell or tissue damage, which is a marker for large number of clinical conditions.

Methodology

Optimized kinetic method of Deutsche. Gessel Schaft fur klinische Chemie (DGKKC).

Principle

Decrease in absorbance due to oxidation of NADH is monitored at 340 nm and directly proportional to LDH activity in the specimen Reagent composition Bring all the reagents (R1 &R2) to room temperature prior to analysis and experiment to be done at 37^{o}C

Assay Procedure

Determine the absorbance change /min (ΔA/Min) for every reading, find the mean avoid time lapsing during experiment .

Calculation

LDH activity (IU /L) = ΔA/Min× T.V×DF
T.V = Total reaction volume in µl
D.F= Dilution factor
S.V=Sample volume µl
Absorptivity = milimolar activity of NADH (6.22mmole/L/cm^{-1}) P= cuvette path length (1 cm)

Estimation of Myeloperoxidase (MPO) [17]

Principle

Myeloperoxidase is an abundant granule heme enzyme which is the most widely studied molecule during last decade which plays a crucial role in inflammation and oxidative stress. MPO assay used for quantitative estimation of intestinal inflammation and oxidative stress. MPO assay used for quantitative estimation of intestinal inflammation due to neutrophil and monocytes infiltration during inflammation and oxidative stress. MPO is copiously expressed in stimulated neutrophil granulocytes, where it catalyses the production of hypoalous acids, such as hypochlorous acid (HOCl), from hydrogen peroxide (H2O2) and chloride ion Cl^{-} or other halides. Myeloperoxidase (MPO) activity was evaluated by the rate of o– dianisidine oxidation measured OD at 460nm.

Procedure

1. **Briefly, inflamed colonic samples of 2 cm length were weighed and homogenized (10% w/v) in an ice cold potassium phosphate buffer (50mM,pH 6.0) which contains Hexadecyl trimethyl ammonium bromide (0.5% w/v) and EDTA (10mM) at 4^{o}c.**

2. **Homogenised samples were subjected to brief period of sonication in an ice cold water bath and freeze thawed three times.**
3. **The samples were sonicated again in an ice water bath and centrifuged at 10000 rpm for a period of 20 minutes.**
4. **100 µl an supernatant was pipette out and mixed with 2.9 ml of phosphate buffer (50mM) contining 0.167 mg/ml of o- dianisidine hydrochloride and 0.0005% hydrogen peroxide (H2O2)**
5. **Immediately, increase in absorbance was recorded every minute for a period of 5 min spectrophotometrically at 460nm**
6. **One unit of MPO activity is defined as the change in absorbance per minute by 1.0 at room temperature, in the final reaction.**
7. **Calculation of MPO activity:**

$$\text{MPO activity (U/g)} = \frac{X}{\text{weight of the piece of tissue taken}}$$

Where,

$$X = 10\times \frac{\text{change in absorbance per min}}{\text{Volume of supernatant taken in the final concentration}}$$

RESULTS

The antiulcer studies were performed on the ethanolic extract of seeds of *Pseudarthria Viscida* Linn. (PVRE) with two doses (200 & 400 mg/kg p.o) and only once administration of the test drug in the experimental rats.

Ethanol (EtOH) induced ulcer Antiulcer studies

The effect of PVRE is shown in table 6 and 7. At dose of 200 & 400 mg/kg it had shown dose dependent decrease in ulcer index. In group I (normal) and in group III (disease control) the ulcer index was shown as 0.83±0.08 and 1.583±0.23 respectively. Group II PVRE Alone (400mg/kg) Group IV PVRE (200 mg/kg + ethanol 1 ml.) and group V PVRE (400 mg/kg + ethanol 1 ml.) group VI (standard) the ulcer index was shown 0.2500±0.11, 0.800±0.12 , 0.700±0.20 and respectively 0.33±0.10 There is decrease in ulcer index when compared with group III. This is tabulated in table 6

The pH of the gastric secretion of group I and group III is 5.26 ± 0.30 , 2.76± 0.27 respectively. There is significant increase in group II, VI, V and VI. 4.25 ± 0.52, 5.11 ± 0.53, 5.56 ± 0.66 and5.38 ± 0.61 respectively compared to group III. Readings are tabulated in table.

The gastric volume of group I and group III is 3.43 ±0.15, 5.06 ± 0.45 respectively. There is a significant decrease in group II, IV, V and V1 the gastric volume was shown as, 2.33±0.18 and 2.43±0.233 respectively compared to group III is observed. This is tabulated in table.

The total acidity of group I (normal) and group III (disease control) 93.39 ± 0.68, 104.7 ±2.013 respectively showed significantly increases of total acidity when compare to normal group. Where as in pretreatment groups II, IV, V and VI groups shows 88.64 ± 4.69, 78.75 ± 2.160, 65.83 ± 2.398 and 72.78 ± 1.989 respectively reduced the total acidity compared to group III. Readings are tabulated in table

PVRE was also studied for its effect on MPO content of the tissue homogenate. It showed rise in MPO level in the control group III (Disease control) treated group , whereas pre-treatment with PVRE at

two doses (200 & 400 mg/kg) there is decline in the MPO levels. Simultaneously, there was a fall in the MPO in Pantoprazole treated group, as shown in (Table-7). There was significant decrease in MPO in the PVRE treated rats at two dose 200 & 400 mg/kg. The MPO content of group I (Normal control) was 3.72 ± 1.18 and group III (Disease Control) 13.06 ± 1.42 showed significantly increased MPO Level, but group II (PVRE alone) ,group IV (PVRE 200 mg/kg), group V (PVRE 400mg/kg) and group VI Pantoprazole (30 mg/kg) , 3.32± 1.23 , 4.46 ± 1.05 , 3.36 ± 0.49 and 3.52 ± 0.76 respectively significantly decreases MPO when compared with ethanol treated group III which is tabulated in table- 6.

PVRE was also studied for its effect on LDH content in the serum. It showed rise in LDH level in the control group III (Disease control) treated group, whereas pre- treatment with PVRE at two doses (200 & 400 mg/kg) there is decline in the LDH levels. Simultaneously, there was a fall in the LDH in Pantoprazole treated group, as shown in (Table-7). There was significant decrease in LDH in the PVRE treated rats at two dose 200 & 400 mg/kg. The MPO content of group I (Normal control) was 10.54 ± 0.97 and group III (Disease Control) 33.59 ± 5.96 showed significantly increased LDH Level, but group II (PVRE alone) ,group IV (PVRE 200 mg/kg), group V (PVRE 400mg/kg) and group VI Pantoprazole (30 mg/kg) , 19.19 ± 2.58 , 20.09 ± 2.74 , 16.17 ± 1.82 and 8.44 ±2.19respectively significantly decreases LDH when compared with ethanol treated group III which is tabulated in table- 6.

Lipid peroxidation estimation is one of the important parameter to access the antiulcer genic effect of PVRE. Lipid peroxidation in group I (normal) was25.32 ± 1.16 and in group III (ethanol 1 ml) lipid peroxidation was 56.37 ± 6.64. In group II, IV, V and VI lipid peroxidation is 26.21±2.60, 37.05 ± 4.79, 31.75± 4.05 and 29.58 ± 4.16, respectively. In pretreated groups there is a significant reduction in lipid peroxidation is observed compared to group III (Table- 7).

Catalase is the most important antioxidant enzyme. In cytoprotection the activity of catalase enzyme is increased. group I (normal) was 27.12 ± 0.80 and In group III (ethanol 1 ml) catalase was 22.61± 0.68 significantly decreased . In group II, IV , V and VI catalase is , 35.06±2.99 , 37.89 ±5.03 , 33.98 ± 3.09 and 37.33 ± 3.38 respectively. In pretreated groups there is a significant reduction in catalase is observed compared to group III (Table- 7).

Histopathological studies are also carried out to access the antiulcer genic effect of PVRE at tissue level. Histopathological studies reveal that destruction of surface epithelium, gastric mucosa and massive infiltration of leucocytes is observed in

Ethanol treated group. It is observed that destruction of gastric mucosa and surface epithelium is significantly reduced in pantaprazole and PVRE treated groups. In some animals of standard and extract treated group infiltration of leucocytes is also observed.

TABLES

Ethanol induced gastric ulcer model

Table 5: - Effect of PVRE on acid volume, pH, ulcer index and total acidity against ethanol induced gastric ulcer in rats.

Sl.no	Group and dose (mg/kg p.o	Acid volume (ml)	pH	Ulcer index	Total acidity (mEq/LI)
1.	Normal Control (Saline)	3.43 ±0.15	5.26 ±0.30	0.83±0.08	93.39 ±0.68
2.	Drug alone (PVRE 400 mg/kg)	3.51 ± 0.30	4.25 ± 0.52	0.2500±0.11	88.64 ±4.69

3.	Disease Control (Absolute ethanol 5ml/ kg)	5.06 ±0.45	2.76± 0.27	1.583±0.23	104.7 ± 2.013
4.	PVRE low dose (200 mg/kg)	3.18± 0.34	5.11 ±0.53	0.800±0.12	78.75 ±2.160
5.	PVRE high dose(400 mg/kg)	2.96 ±0.14	5.56 ±0.66	0.700±0.20	65.83 ±2.398
6.	Pantaprazole (30 mg/kg)	2.86 ± 0.18	5.38 ±0.61	0.33±0.10	72.78 ± 1.989

Values are expressed as mean ± SEM of 6 animals in each group. Data analysed by One way ANOVA followed by Tukey's test Significant relative to a= P < 0.001, b=P < 0.01, c= P <0.05 when compared with control group.

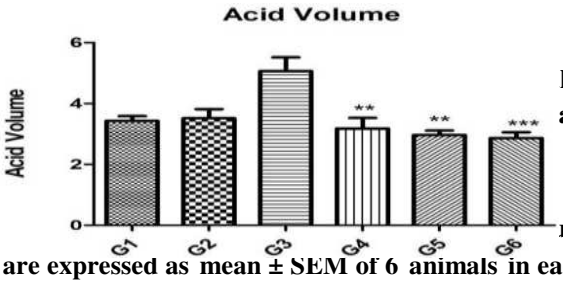
Table5.1 :-Effect of PVRE on LDH, MPO, lipid peroxidation and catalase against ethanol induced gastric ulcer in rats.

Sl.no	Group and dose (mg/kg p.o	MPO	LDH	LPO (nM/mgof protein)	Catalase units/mg of protein
1.	Normal Control (Saline)	3.72 ± 1.18	10.54 ±0.97	25.32 ±1.16	27.12 ± 0.80
2.	Drug alone (PVRE 400 mg/kg)	3.32±1.23	19.19 ± 2.58	26.21± 2.60	35.06 ±2.99
3.	Disease Control (Absolute ethanol 5 ml/ kg)	13.06 ± 1.42	33.59 ± 5.96	56.37 ±6.64	22.61±0.68
4.	PVRE low dose (200 mg/kg)	4.46 ±1.05	20.09 ± 2.74	37.05 ±4.79	37.89 ±5.03
5.	PVRE high dose(400 mg/kg)	3.36 ±0.49	16.17 ±1.82	31.75± 4.05	33.98 ± 3.09
6.	Pantaprazole (30 mg/kg)	3.52 ±0.76	8.44 ± 2.19	29.58 ± 4.16	37.33 ± 3.38

Values are expressed as mean ± SEM of 6 animals in each group. Data analysed by One way ANOVA followed by Tukey's test Significant relative to a= P < 0.001, b=P < 0.01, c= P <0.05 when compared with control group.

Figure 17: - Effect of PVRE on acid volume using in different groups in ethanol induced gastric ulcer model.

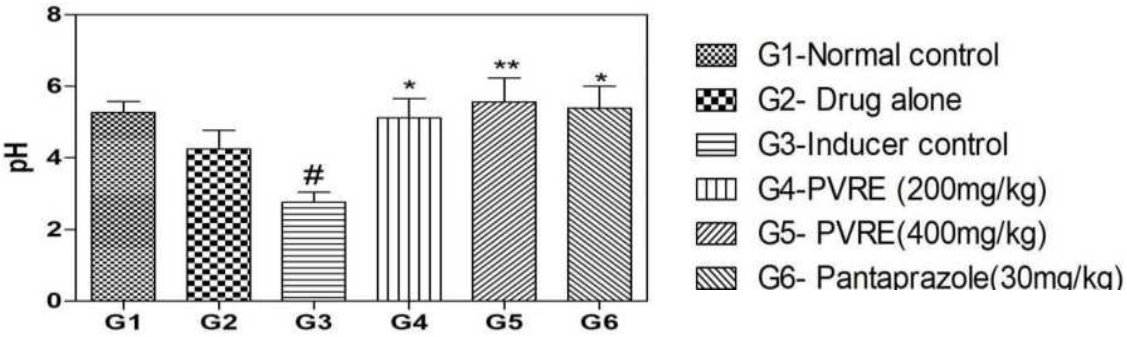




Valu ls in each group. Data analysed by One- w a y
AN ative to P < 0.001P <0.05 when compared with
con

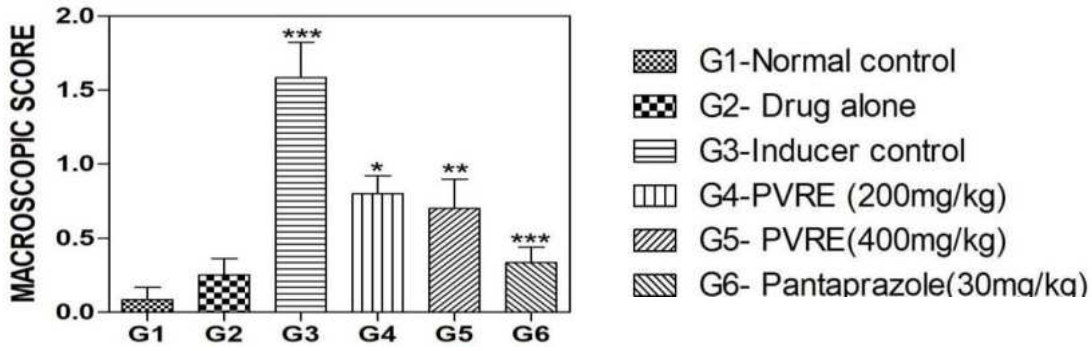
Figure 1 ips in ethanol induced gastric ulcer model.

Values are expressed as mean ± SEM of 6 animals in each group. Data analysed by One way ANOVA followed by



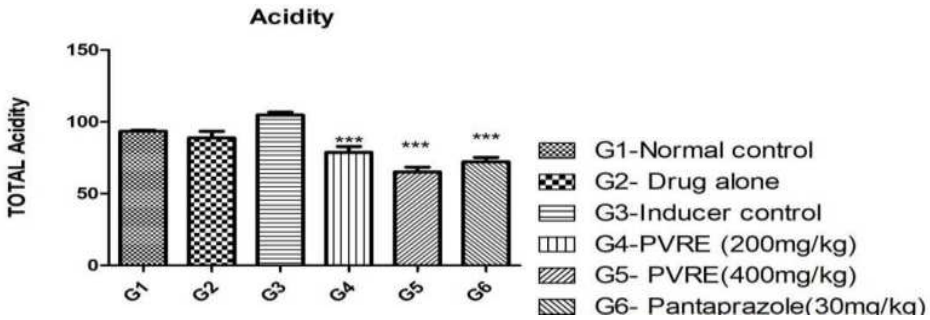
Tukey's test Significant relative to P < 0.001, P<0.05 when compared with control group

Figure 19:- Effect of PVRE on ulcer index using in diffferent groups in ethanol induced gastric ulcer model.



Values are expressed as mean ± SEM of 6 animals in each group. Data analysed by One way ANOVA followed by Tukey's test Significant relative to P < 0.001, P <0.05 when compared with control group

Figure 20:- Effect of PVRE on total acidity using in different groups in ethanol induced gastric ulcer model.



Values are expressed as mean ± SEM of 6 animals in each group. Data analysed by One way ANOVA followed by Tukey's test Significant relative to P < 0.001, P <0.05 when compared with control group

Figure 21:- Effect of PVRE on MPO using in diffferent groups in ethanol induced gastric ulcer model.

Values are expressed as mean ± SEM of 6 animals in each group. Data analysed by One way ANOVA followed by Tukey's test Significant relative to P < 0.001,P<0.05 when compared with control group



Figure 22:- Effect of PVRE on LDH using in diffferent groups in ethanol induced gastric ulcer model.

Values are expressed as mean ± SEM of 6 animals in each group. Data analysed by One-way ANOVA followed by Tukey's test Significant relative to P < 0.001, P <0.05 when compared with control group.

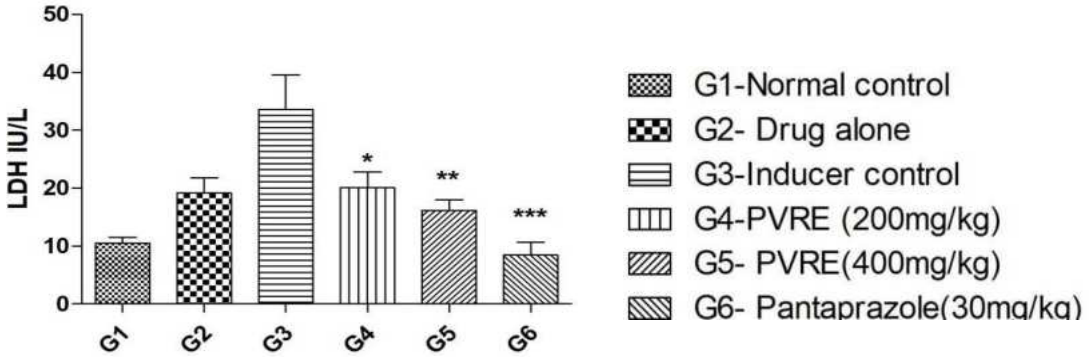
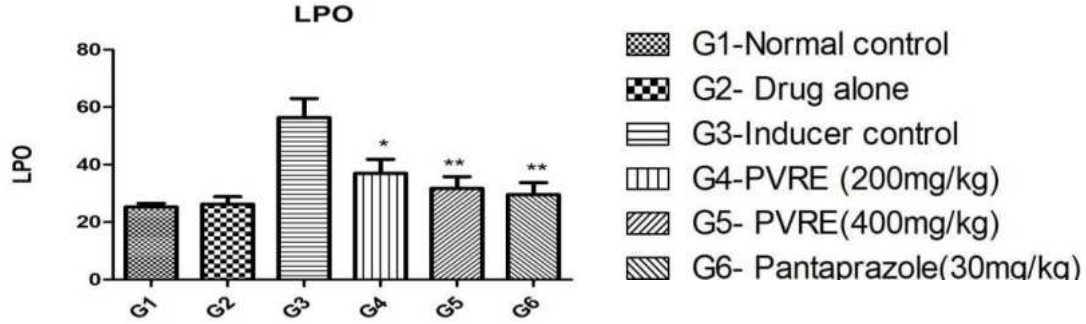
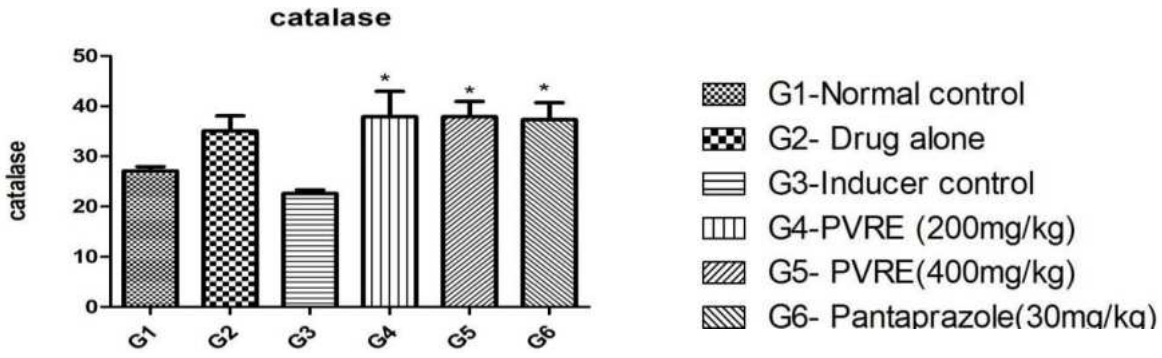


Figure 23:- Effect of PVRE on LPO using in diffferent groups in ethanol induced gastric ulcer model.



Values are expressed as mean ± SEM of 6 animals in each group. Data analysed by One-way ANOVA followed by Tukey's test Significant relative to P < 0.001, P<0.05 when compared with control group

Figure 24:- Effect of PVRE on catalase using in diffferent groups in ethanol induced gastric ulcer model.



Values are expressed as mean ± SEM of 6 animals in each group. Data analysed by One-way ANOVA followed by Tukey's test Significant relative to P < 0.001, P<0.05 when compared with control group

Figure 25:- Stomachs of diffenrent groups of rats showing gastric ulcer induced by ethanol.

Normal Group



Inducer Group



PVRE Low dose (200 mg/kg)



PVRE High dose (400 mg/kg)

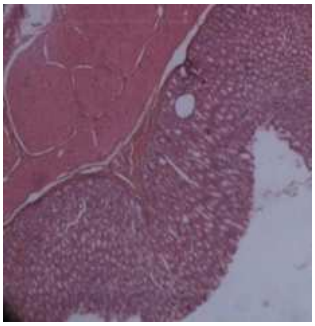


Drug Alone PVRE(400mg/kg)

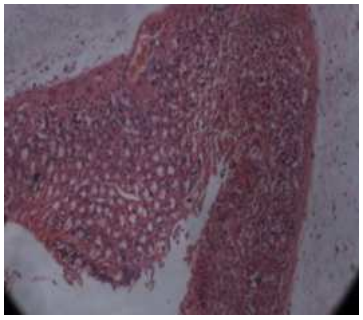


Pantoprazole (30mg/kg)

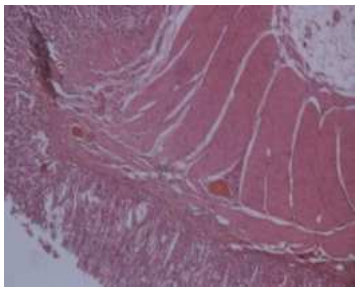
Figure 26:- Histopathogical analysis of rat stomachs in ethanol induced ulcer model



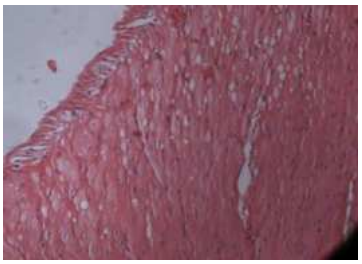
Normal Group



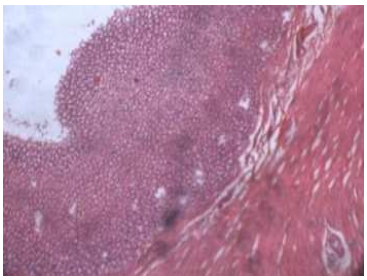
Inducer Group



PVRE High dose(400mg/kg)



PVRE low dose(200mg/kg)



Pantoprazole(30mg/kg)

Indomethacin induced gastric ulcer model. Antiulcer studies

The effect of PVRE is shown in table 6 and 7. At dose of 200 & 400 mg/kg it had shown dose dependent decrease in ulcer index. In group I (normal) and in group III (disease control) the ulcer index was shown as 0.97±0.05 and 3.17±0.18 respectively. Group II PVRE Alone (400mg/kg) Group IV PVRE (200 mg/kg + indo (50mg/kg) and group V PVRE (400 mg/kg + indo (50mg/kg) group VI (standard) the ulcer index was shown 1.19±0.118, 1.43±0.18 , 1.42±0.21 and respectively 1.58±0.29 There is decrease in ulcer index when compared with group III. This is tabulated in table 6.

The pH of the gastric secretion of group I and group III is 4.5 ± 0.19 , 2.76± 0.28 respectively. There is significant increase in group II, VI, V and VI. 4.23 ± 0.35, 5.93 ± 0.98, 5.93± 0.98 and 4.63 ± 0.311 respectively compared to group III. Readings are tabulated in table.

The gastric volume of group I and group III is 3.60 ±0.18, 5.33 ± 0.46 respectively. There is a significant decrease in group II, IV, V and V1 the gastric volume was shown as, 4.54±0.51, 3.36 ± 0.22, 3.37 ± 0.34 and 3.14±0.34 respectively compared to group III is observed. This is tabulated in table

The total acidity of group I (normal) and group III (disease control) 97.04 ± 0.62, 105.0 ±1.54 respectively showed significantly increases of total acidity when compare to normal group. Where as in pretreatment groups II, IV, V and VI groups shows 89.30 ± 2.87, 86.00 ± 1.733, 79.56 ± 2.22 and 83.03 ± 3.970 respectively reduced the total acidity compared to group III. Readings are tabulated in table

PVRE was also studied for its effect on MPO content of the tissue homogenate. It showed rise in MPO level in the control group III (Disease control) treated group, whereas pre-treatment with PVRE at two doses (200 & 400 mg/kg) there is decline in the MPO levels. Simultaneously, there was a fall in the MPO in Pantoprazole treated group, as shown in (Table-7). There was significant decrease in MPO in the PVRE treated rats at two dose 200 & 400 mg/kg. The MPO content of group I (Normal control) was 3.54 ± 1.15 and group III (Disease Control) 13.12 ± 1.39 showed significantly increased MPO Level, but group II (PVRE alone), group IV (PVRE 200 mg/kg), group V (PVRE 400mg/kg) and group VI Pantoprazole (30 mg/kg), 3.28 ,1.09, 4.36 ± 0.95, 3.32 ± 0.37 and 4.58 ± 1.09 respectively significantly decreases MPO when compared with indomethacin treated group III which is tabulated in table

PVRE was also studied for its effect on LDH content in the serum. It showed rise in LDH level in the control group III (Disease control) treated group, whereas pre- treatment with PVRE at two doses (200 & 400 mg/kg) there is decline in the LDH levels. Simultaneously, there was a fall in the LDH in Pantoprazole treated group, as shown in (Table-7). There was significant decrease in LDH in the PVRE treated rats at two dose 200 & 400 mg/kg. The MPO content of group I (Normal control) was 10.42 ± 0.58 and group III (Disease Control) 39.10 ± 3.95 showed significantly increased LDH Level, but group II (PVRE alone), group IV (PVRE 200mg/kg), group V (PVRE 400mg/kg) and group VI Pantoprazole (30 mg/kg), 20.30 ± 2.04 ,13.0 ± 2.004, 17.36± 1.37 and 21±6.82 respectively significantly decreases LDH when compared with indomethacin treated group III which is tabulated in table- 6.

Lipid peroxidation estimation is one of the important parameter to access the antiulcer genic effect of PVRE. Lipid peroxidation in group I (normal) was24.85 ± 0.45 and in group III (Indo

50 mg/kg) lipid peroxidation was 61.35± 1.62. In group II, IV, V and VI lipid peroxidation is 26.37±1.12, 36.13 ±1.92, 27.82± 2.09 and 28.58 ± 1.83, respectively. In pretreated groups there is a significant reduction in lipid peroxidation is observed compared to group III (Table- 7).

Catalase is the most important antioxidant enzyme. In cytoprotection the activity of catalase enzyme is increased. group I (normal) was 29.17 ± 0.47 and in group III (Indo 50 mg/kg) catalase was 23.36± 1.036 significantly decreased. In group II, IV, V and VI catalase is, 34.74±1.66, 31.66 ±2.06, 33.37 ± 2.58 and 33.52 ± 1.539 respectively. In pretreated groups there is a significant reduction in catalase is observed compared to group III (Table- 7).

Histopathological studies are also carried out to access the antiulcer genic effect of PVRE at tissue level. Histopathological studies reveal that destruction of surface epithelium, gastric mucosa and massive infiltration of leucocytes is observd.

Indomethacin treated group. It is observed that destruction of gastric mucosa and surface epithelium is significantly reduced in Pantaprazole and PVRE treated groups. In some animals of standard and extract treated group infiltration of leucocytes is also observed.

TABLES

Indomethacin induced gastric ulcer model

Table 6:- Effect of PVRE on acid volume, pH, ulcer index and total acidity against Indomethacin induced gastric ulcer in rats.

Sl.no	Group and dose (mg/kg p.o	Acid volume (ml)	pH	Ulcer index	Totalacidity (mEq/LI)
1.	Normal Control (Saline)	3.60±0.18	4.5 ±0.19	0.97 ±0.05	97.04 ±0.62
2.	Drug alone (PVRE400 mg/kg)	4.54 ±0.51	4.23 ±0.35	1.19 ±0.118	89.30 ± 2.87
3.	Disease Control (Indomethacin 50mg/kg)	5.33 ±0.46	2.76 ± 0.28	3.17 ± 0.18	105.0 ± 1.54
4.	PVRE low dose (200 mg/kg)	3.36 ±0.22	5.05 ± 0.39	1.43 ±0.18	86.00 ±1.733
5.	PVRE high dose(400 mg/kg)	3.14 ±0.34	5.93± 0.98	1.42 ±0.21	79.56 ± 2.22
6.	Pantaprazole (30 mg/kg)	3.37 ±0.32	4.63 ±0.311	1.58 ± 0.29	83.03 ± 3.970

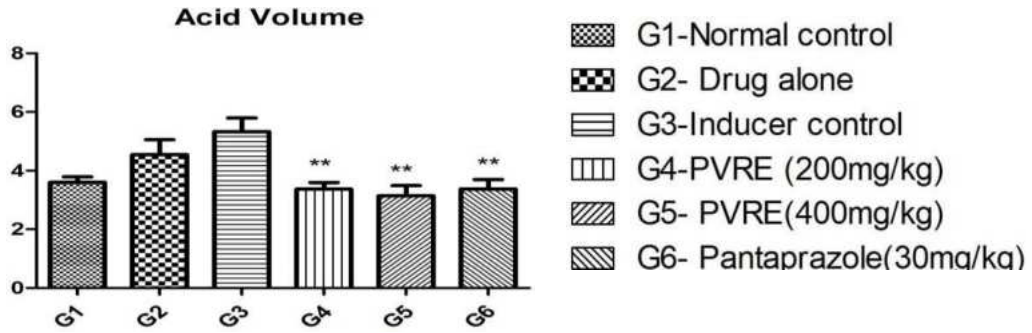
Values are expressed as mean ± SEM of 6 animals in each group. Data analysed by One way ANOVA followed by Tukey's test Significant relative to a= P < 0.001, b=P< 0.01, c= P <0.05 when compared with control group.

Table 6.1:-Effect of PVRE on LDH, MPO, lipid peroxidation and catalase against Indomethacin induced gastric ulcer in rats.

Sl.no	Group and dose (mg/kg p.o	MPO	LDH	LPO (nM/mg of protein)	Catalase units/mg of protein
1.	Normal Control (Saline)	3.54 ±1.15	10.42 ± 0.58	24.85 ±0.45	29.17 ± 0.47
2.	Drug alone (PVRE 400 mg/kg)	3.28 ±1.09	20.30± 2.04	26.37 ±1.12	34.74 ±1.66
3.	Disease Control (Indomethacin 50mg/kg)	13.12 ±1.39	39.10 ±3.95	61.35 ± 1.62	23.36 ± 1.036
4.	PVRE low dose (200 mg/kg)	4.36 ± 0.95	13. ±2.004	36.13 ±1.92	31.66 ± 2.06
5.	PVRE high dose(400 mg/kg)	3.32 ± 0.37	17.36 ±1.37	27.82 ±2.09	33.37 ±2.58
6.	Pantaprazole (30 mg/kg)	4.58 ± 1.09	21.40 ±6.82	28.58 ± 1.83	33.52 ±1.539

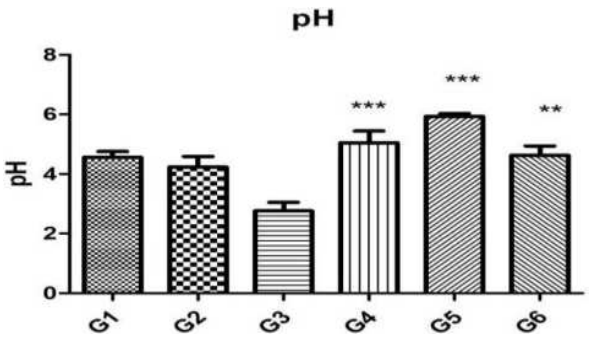
Values are expressed as mean ± SEM of 6 animals in each group. Data analysed by One way ANOVA followed by Tukey's test Significant relative to a= P < 0.001, b=P < 0.01, c= P <0.05 when compared with control group.

Fig 27:- Effect of PVRE on Acid volume using in diffferent groups in Indomethacin induced gastric ulcer.

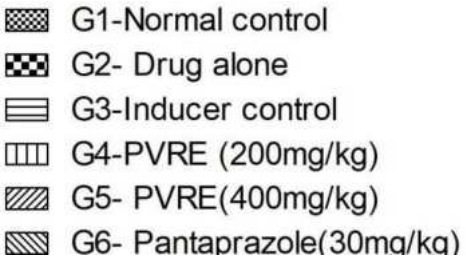


Values are expressed as mean ± SEM of 6 animals in each group. Data analysed by One way ANOVAfollowedbyTukey'stestSignificantrelativetoP<0.001,P<0.05 when compared with control group

Fig 28:- Effect of PVRE on pH using in diffferent groups in Indomethacin induced gastric ulcer



model.

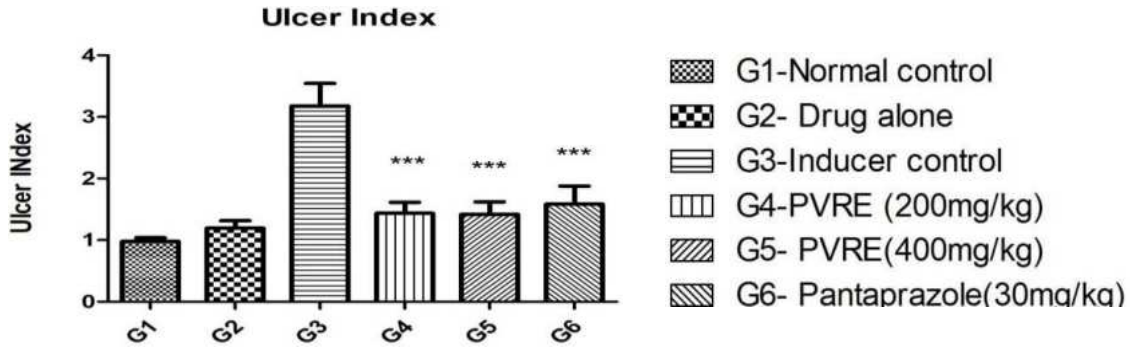


Values are expressed as mean ± SEM of 6 animals in each group. Data analysed by One way ANOVA followed

by Tukey's test Significant relative to P < 0.001, P<0.05 when compared with control group

Fig 29:- Effect of PVRE on Ulcer index using in diffferent groups in Indomethacin induced

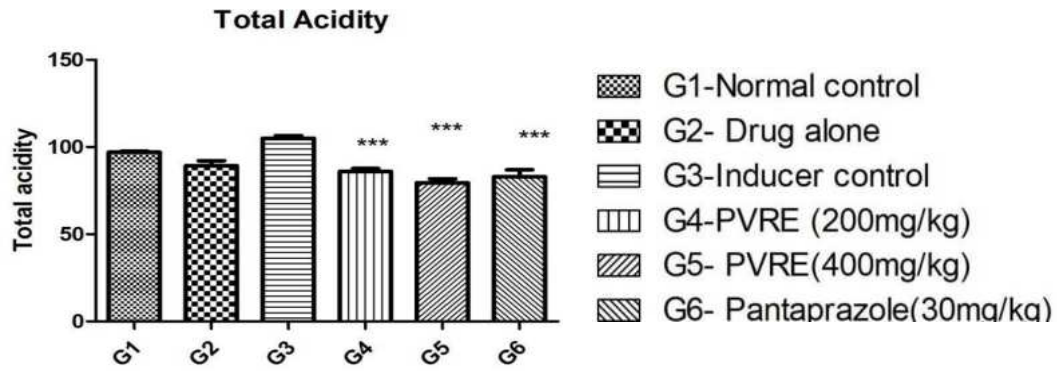
gastric ulcer model.



Values are expressed as mean ± SEM of 6 animals in each group. Data analysed by One way ANOVA followed by Tukey's test Significant relative to P < 0.001,P <0.05 when compared with control group

Fig 30:- Effect of PVRE on Total acidity using in diffferent groups in Indomethacin induced

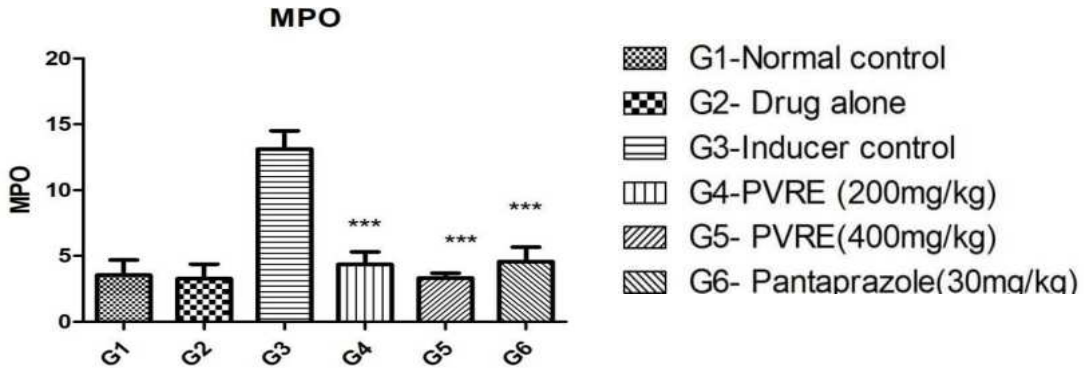
gastric ulcer model.



Values are expressed as mean ± SEM of 6 animals in each group. Data analysed by One way ANOVA followed

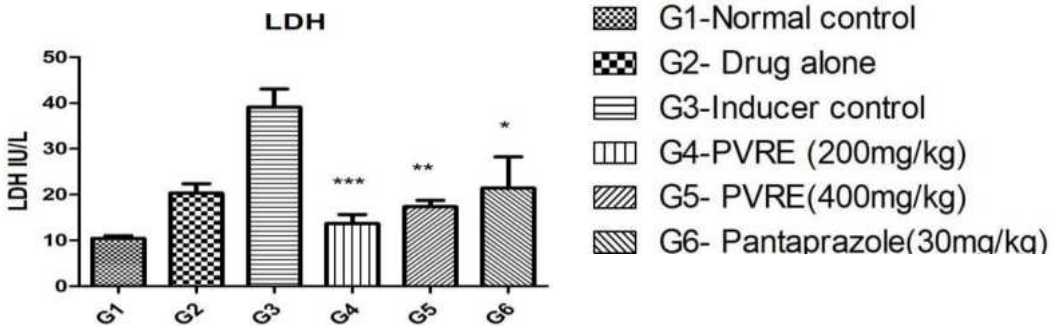
by Tukey's test Significant relative to P < 0.001, P <0.05 when compared with control group

Fig 31:- Effect of PVRE on MPO using in diffferent groups in Indomethacin induced gastric ulcer model.



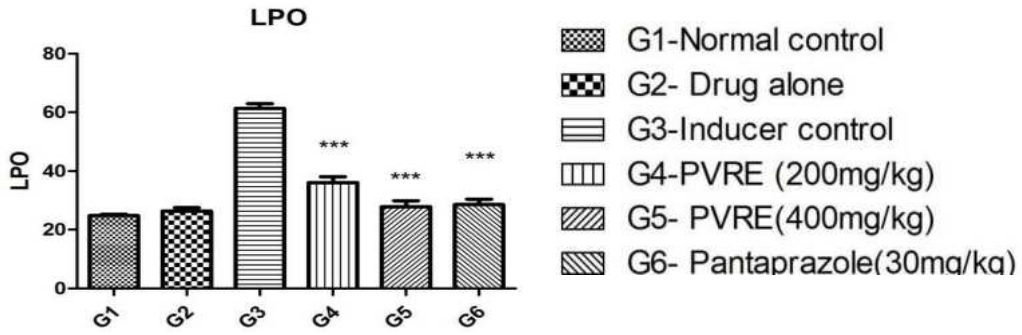
Values are expressed as mean ± SEM of 6 animals in each group. Data analysed by One way ANOVA followedbyTukey'stestSignificantrelativetoP<0.001 P<0.05 when comp

Fig 32:- Effect of PVRE on LDH using in diffferent groups in Indomethacin induce gastric ulcer model



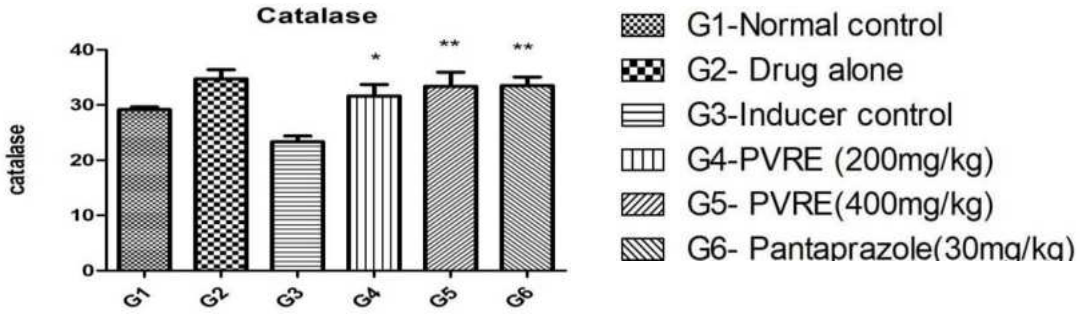
Values are expressed as mean ± SEM of 6 animals in each group. Data analysed by One way ANOVA followed by Tukey's test Significant relative to P < 0.001,P<0.05 when compared with control group

Fig 33:- Effect of PVRE on lipid peroxidase using in diffferent groups in Indomethacin induced gastric ulcer model.



Values are expressed as mean ± SEM of 6 animals in each group. Data analysed by One way ANOVA followed by Tukey's test Significant relative to P < 0.001, P <0.05 when compared with control group

Fig 34:- Effect of PVRE on Catalase using in diffferent groups in Indomethacin induced gastric ulcer model



Values are expressed as mean ± SEM of 6 animals in each group. Data analysed by One-way ANOVA followed by Tukey's test Significant relative to P<0.001, P <0.05 when compared with control group.

FIGURE 35:- Stomachs of diffenrent groups of rats showing gastriculcer induced by indomethacin.

Normal Control



InducerControl



PVRE Low dose (200mg/kg)



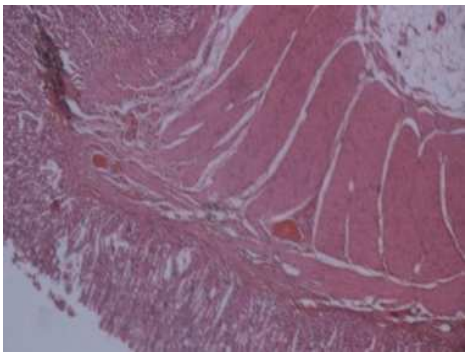
PVRE high dose (400 mg/ kg)

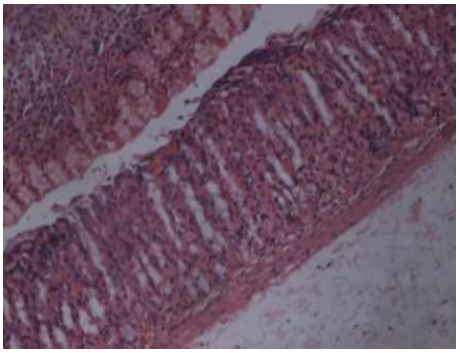


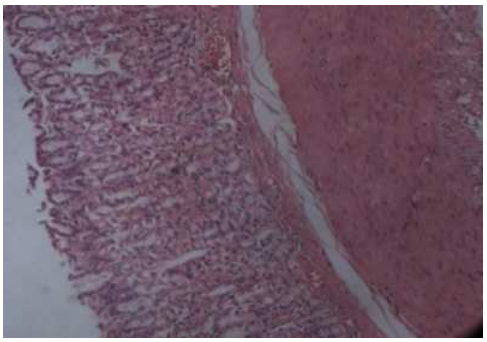
Pantoprazole (30 mg/kg)



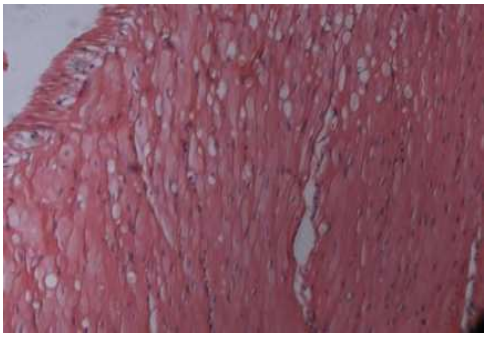
FIGURE 36:- Histopathogical analysis of rat stomachs in Indomethacin induced ulcer model







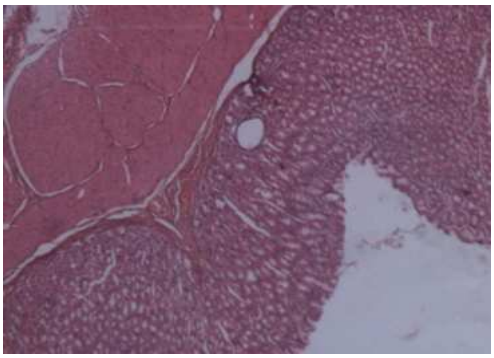
Normal Control



Inducer Control

PVRE low dose(200 mg/kg)

PVRE high dose (400 mg/kg)



Pantoprazole (30 mg/kg)

DISCUSSION

Gastric and duodenal ulcers are illnesses that affect a considerable number of people in the world and some authors consider gastric ulcer as the new plague of the 21st century[18]. Factors such as stress, smoking, nutritional deficiencies and frequent ingestion of nonsteroidal-inflammmatory drugs (NSAIDS) grow the gastric ulcer incidence. In general, it is caused by an imbalance between aggressive and defensive factors of the gastric mucosa.

The modern approach to control gastro duodenal ulceration is to scavenge reactive oxygen species, inhibiting H^+k^+ ATPase pump to control increased acid secretion and eradication of *H.Pylori*. A range of drugs like histamine blockers and proton pump inhibitors although has been used for efficient management of gastric hyper secretion, many of these drugs pose adverse effects like dizziness, drowsiness, gas accumulation, headache, nausea, vomiting, inflammation of the nose, etc[12.]

Traditional medicine made use of plants since ancient times to treat varied gastrointestinal diseases including peptic ulcers. Numerous studies have been published involving tests with medicinal plants showing antiulcer activity[19]. Diverse chemical compounds have been isolated from medicinal plants with antiulcer activity. This is an important reason to investigate antiulcer effects in medicinal plants with traditional use in gastric diseases[20].

The present study is carried out to investigate the antiulcer activity of Hydroalcoholicextract of *Pseudarthria viscida* root extract in ethanol induced and indomethacin induced gastric ulcers. Two doses of PVRE 200 and 400 mg/kg are used in present ulcer index, acid gastric volume, pH, total acidity, MPO, LDH, lipid peroxidase and catalase estimation is carried out in assessment of antiulcer activity.

The present study, Pantoprazole used as a standard drug. Pantoprazole is a Proton pump inhibitor, is capable of inhibiting the final common step in gastric acid secretion. pantoprazole is inactive at neutral pH , but at pH less than 5 it rearranges to two charged cationic forms (a sulphenic acid and a sulphenamide configurations) that react covalently with SH groups of the H^+k^+ ATPase enzyme and inactivate it irreversibly, especially when two molecules of omeprazole react with one molecules of the enzymes After absorption into blood stream and subsequent diffusion into the parietal cell, it gets concentrated in the acidic pH of the canaliculi because the charged forms generated there are unable to diffuse back. Moreover, it gets tightly bound to the enzymes by covalent bonds. These features and the specific localization of H^+k^+ ATPase to the apical membrane of the parietal cells confer high degree of selectivity of action to pantoprazole Acid secretion resumes only when H^+k^+ ATPase molecules are synthesized. It also inhibits gastric mucosal carbonic anhydrase.

In the present study the hydro alcoholic extract of *Pseudarthria Viscida* Linn. At dose levels of 200 and 400 mg/kg marked gastro protection in two models, i.e. Ethanol, indomethacin showing more protection. The efficacy of PVRE over Pantaprazole is observed in both models.

In ethanol and indomethacin induced gastric ulcer model significantly reduced ulcer index, acid volume, total acidity, total protein content and lipid peroxidase. It also caused significant increase in pH, catalase and glutathione. Pantoprazole shown better activity compared to extract treated groups.

Histopathological studies also confirmed that pre-treatment with PVRE inhibited the ethanol and indomethacin induced congestion, hemorrhage and necrosis in gastric mucosa.

Ethanol has been shown to produce free radicals and induce peptic ulcers. While stress ulcers are mediated by brain gut axis and complex neural mechanism. Stress causes an ischemic

condition in the gastric mucosa by the activation of the parasympathetic and sympathetic nervous system resulting in vasoconstriction, which in turn causes free radical generation. Further, stress has also found to inactivate mucosal prostaglandins known to favor the generation H2O2, which in turn inhibits the synthesis of prostaglandins known to favor the generation of reactive oxygen species. This in turn increases the influx of Ca^{2+} ions, resulting in reduced membrane integrity of surface epithelial cells, thereby generating gastric ulcers[21].

Indomethacin is a potent prostaglandin (PG) biosynthesis inhibitor, and inhibition of PG synthesis by indomethacin coincides with the earlier stages of damage to the cell membranes of mucosal, parietal and endothelial cells. It has been reported that gastric acid secretion is involved in the formation of indomethacin-induced mucosal lesions. The PVRE significantly reduced the ulcer index and afforded significant protection against indomethacin-induced gastric ulcers.

From the above discussion it is evident that the PVRE pre-treatment in both ulcer inducing models, i.e. indomethacin, ethanol caused significant reduction in ulcer severity. In both gastric ulcers model the antiulcer activity of PVRE was better than the standard drug used in present study. Based on the significant reduction in ulcer lesions in all both models mentioned above it can be concluded that PVRE lowers the risk and incidence of ulcers as well as helps in treatment of active ulcers.

CONCLUSION

The study has been done to investigate the antiulcer activity of PVRE in ethanol and indomethacin induced gastric ulcer. It has been found that the PVRE possess significant antiulcer activity in dose dependent manner. It has been observed that in both models there is decrease in acid volume, ulcer index, total acidity, LDH, MPO, lipid peroxidase and increase in pH and catalase,compared to control group. Histopathological studies also confirmed that PVRE has Cytoprotective effect.

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